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Composts and Organic By-Products in *Pinus halepensis* Forestry

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In the sustainable organic waste management context, this study was carried out to explore the potential of easily available composts and organic by-products for *Pinus halepensis* forestry in Mediterranean areas. The study consisted in four experiments using locally abundant organic waste: experiments I and IV evaluated their suitability as fertilizers for nurseries and pine replanting respectively; experiment II considered vermicompost efficiency as a root-growth promoter; experiment III assessed the aptitude of organic waste as a component of forestry substrates. Different plant measurements, morphological attributes and analytical determinations were used to assess seedling quality and to evaluate substrates and fertilizers' aptitude. Fertilization experiments run under nursery conditions showed that organic waste produced plantlets with similar morphological and nutritional states to mineral products with good quality standards. In pine replanting, organic fertilization clearly increased soil organic matter levels, enhancing soil nutritional and structural conditions. When evaluating different mixtures as substrate, no large differences appeared among treatments, except for the seedlings grown in virgin or recycled coconut fiber, where greater aerial development was achieved. Further research is needed to comprehend why the use of vermicompost as a root-growth promoter did not produce the expected effects. The obtained results indicate that organic by-products and composts can substitute the mineral fertilizers and substrates usually employed in the Mediterranean Region in *P. halepensis* forestry to contribute to develop a local circular economy.

Keywords: vermicompost, alternative growing media, root-growth promoter, seedling growth and nutrition, reforestation

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INTRODUCTION

Pinus halepensis is widely used for reforestation and urban gardening in the Mediterranean Region because it well adapts to poor soil, high temperatures, and to scarce irregular rainfall. This region is often severely affected by agricultural transformation, mismanaged wood exploitation and frequent fires. Its reforestation is encouraged by European Union authorities in order to reduce environmental degradation and preserve soil quality to, in turn, lead to a high demand of plantlets for the nurseries. Large amounts of customized substrates containing non-renewable materials, such as peat or natural soil, are used, which simply add to the list of environmental problems.

However, the ecosystem damage caused by soil and peat extraction can be avoided if these materials are fully or partially replaced with organic materials that possess comparable porosity and aeration properties (Ostos et al., 2008). There is a wide array of available waste and by-products with different characteristics that can be used or combined to produce substrates: composts made from municipal refuse or green waste, sewage sludge, pine bark, sawmill residue, grape marc, rice hull and many other materials have been successfully used as components for substrates that are suited for many different plants. In forestry nurseries, Veijalainen et al. (2008) and Lilja et al. (2013) showed the potential of on-site biodegradable waste (discarded plants, exhausted substrates, and other vegetal residue such as weeds, grasses and fallen leaves) to be used as minor components in growing media. Similarly, many other types of organic materials, such as activated sewage sludge, composted sewage sludge, municipal solid waste and paper mill sludge, have complemented peat or pine bark in substrates to grow *Cupressus arizonica, C. sempervirens*, *Eucaliptus grandis, Pinus halepensis*, *P. pinaster*, *P. pinea*, and *Pistacia lentiscus* (Guerrero et al., 2002; Hernández-Apaloaza et al., 2005; Ostos et al., 2008; Mañas et al., 2009, 2010; Toledo et al., 2015).

Nevertheless, organic materials can be used in forestry in more ways than merely as substrate components. Some are routinely used as organic fertilizers in field plantations, but have also had beneficial effects in nurseries. The application of chicken manure or green compost, for instance, improves the morphological quality of *Fraxinus pennsylvanica* and *Quercus rubra* seedlings (Davis et al., 2006). Similarly, chicken manure amendments combined with inorganic fertilization in *Larix olgensis* seedlings increase post-transplant new root numbers (Wei and Guo, 2016). Even though the less sophisticated types are the most commonly used because they are low-cost, high-value products, such as vermicompost, organic fertilizer produced under mesophilic conditions by the joint interaction of earthworms and microorganisms in organic matter breakdown, have found their place in forestry. Its beneficial effects range from improving physical substrate properties to acting as a source of nutrients, together with providing plant growth regulators, such as fulvic and humic acids, which have been reported to increase the number of roots (Bachman and Metzger, 2008; Aremu et al., 2012). In forestry, vermicompost has been demonstrated as a stimulator of germination and growth for species like *Licania tomentosa* (Alves and Passoni, 1997), *Eucalyptus* spp. (Kandari et al., 2011), *Leucaena leucocephala* and *Cassia fistula* (Madan et al., 2009), *Pterocarpus marsupium* (Venkatesh et al., 2009), and *P. pinaster* (Lazcano et al., 2010).

Recycling organic waste in forestry and agriculture is an important strategy for circular economy, whose priority prevails in the EU. Properties of all organic residue and derived products show wide local and temporal variabilities. The edaphoclimatic characteristics of an area, together with market interests, will decide the varieties to be planted and how they will be managed. So, local conditions should always be taken into account when considering an optimal organic residue valorization in sustainable management (Hicklenton et al., 2001; Hernández-Apaloaza et al., 2005; Ostos et al., 2008; Lazcano et al., 2010).

The aim of this study was to explore the potential uses (fertilizers for nurseries and forest replanting, root-growth promoters and substrate components) of easily available composts and organic by-products and residues for *Pinus halepensis* forestry in the Mediterranean Region. Four different trials were carried out, and special emphasis was placed to adapt their objectives and design to the sector's needs, use patterns and the typical characteristics of the tested organic materials.

MATERIALS AND METHODS

Experimental Design

The study was carried out using locally available organic sources: sewage sludge compost, meat bone and meal, municipal solid waste compost, ovine manure, two refuses from significant olive oil local production and coconut fiber, previously used in greenhouse vegetable production, a widespread activity in the study area. Vermicompost from municipal solid waste was also used to evaluate its capacity to increase the root system of plantlets by contributing to the future resistance of trees under Mediterranean conditions, which are usually stressing. **Table 1** displays the analytical properties of the tested organic matters. The abbreviations employed for each organic product throughout the study are as follows:

SSC: Sewage Sludge Compost
MBM: Meat Bone and Meal
VC: Vermicompost
CF: Coconut Fiber
RCF: Recycled Coconut Fiber
MSWC: Municipal Solid Waste Compost
GOS: Ground Olive Stone
OM: Ovine Manure
OMPC: Olive Mill Pomace Compost

According to the potential uses considered herein, four different pot experiments using *P. halepensis* plantlets were conducted in an open-air plot at the experimental farm of the Valencian Institute of Agricultural Research in Moncada (39° 33′ N; 24° 24′ W, Valencia, Spain). The meteorological conditions, provided by the nearest pluviometric observatory, were: annual average precipitation of 451 mm, annual average temperature of 15.4°C and the annual average potential evapotranspiration was 629 mm. Plants were periodically watered to maintain optimal humidity. All the experiments shared a randomized-block design and a similar setup, and differed only in terms of the treatments evaluated and some minor details needed to adapt them to meet their objectives. The substrate used in experiments I, II and III was a commercial substrate provided for the collaborating nursery, made of forest soil, palm tree pruning, pine pruning and coconut fiber (bulk density: 0.467 g cm^3; water-holding capacity: 54.5%; pH in substrate: water dilution 1:25: 9.19; electric conductivity in substrate: water dilution 1:5: 2.35 dS m^{-1}). The experiments were as follows:

Experiment I: Evaluation of Sewage Sludge Compost and Meat and Bone Meal as a Fertilizer for Nurseries

Fifty homogeneous pine plantlets, about 10 cm high, were grown individually in 3-liter plastic pots filled with commercial substrate. Five fertilizer treatments (10 replicates each) were compared: C (slow-release NPK 16:8:12 fertilizer, Basacote Plus

TABLE 1 | Chemical characteristics of organic refuses.

	SSC	MBM	VC	CF	RCF	MSWC	OM	OMPC
OM_ox. (g Kg^{-1} dw)	650	681	383	541	636	676	429	681
Total N (g Kg^{-1} dw)	28.7	76.6	22.0	4.00	16.7	19.2	23.9	28.2
C/N	13.1	5.16	10.1	78.5	22.1	20.2	10.4	14.0
P_2O_5 (g Kg^{-1} dw)	18.9	93.2	41.3	0.80	3.62	9.60	25.2	23.6
K_2O (g Kg^{-1} dw)	2.00	8.60	2.20	10.8	2.30	8.20	28.9	47.6
CaO (g Kg^{-1} dw)	60.4	137	133	3.00	6.53	76.1	109	32.6
MgO (g Kg^{-1} dw)	4.60	4.00	11.0	2.10	9.30	8.30	14.0	16.3
Na_2O (g Kg^{-1} dw)	1.10	8.00	0.30	3.90	4.50	6.40	8.50	5.90
pH (aquose sol. 1:25)	7.38	6.57	8.65	5.44	6.18	6.39	8.70	9.38
EC(aquose sol. 1:5, dSm^{-1})	5.10	6.77	4.29	3.73	4.41	13.5	12.6	16.8

OM_ox., oxidable organic matter; dw, dry weight; EC, electrical conductivity.

SM-Compo Expert Spain at the rate of 0.5 g N l^{-1}), SSC1 (SSC at the rate of 0.5 g N l^{-1}), SSC2 (SSC at the rate of 1 g N l^{-1}), MBM1 (MBM at the rate of 0.5 g N l^{-1}), MBM2 (MBM at the rate of 1 g N l^{-1}).

Experiment II: Evaluation of Vermicompost as a Root-Growth Promoter for Pine Plantlets

Sixty homogeneous pine plantlets, around 10 cm high, were grown individually in 3-liter plastic pots filled with commercial substrate. Six treatments (10 replicates each) were tested: C (non-amended control) and VC1 to VC5 (5, 10, 20, 40, and 80 g of vermicompost per pot in fresh weight, respectively). All the plants were also fertilized with a slow-release NPK (16:8:12) fertilizer, Basacote Plus SM Compo Expert Spain at the rate of 0.5 g N l^{-1}. Vermicompost was produced by the earthworm *Eisenia fetida* from MSWC in containers of 600 L. MSWC (300 kg) was inoculated with 2000 adult worms (*Eisenia foetida*), keeping its moisture content between 60 and 70%.

Experiment III: Evaluation of Coconut Fiber, Recycled Coconut Fiber, Municipal Solid Waste Compost and Ground Olive Stone as Forestry Substrate Components

Using 3-liter plastic pots and pine plantlets about 10 cm high, eight different substrates were tested (10 replicates per treatment): C (customized commercial substrate), CF, RCF, RCF/MSWC 2:1, RCF/MSWC 3:1, RCF/GOS 2:1, RCF/GOS 3:1, and RCF/MSWC/GOS 2:1:1. All the mixing ratios are expressed as volume. All the plants were fertilized with a slow-release NPK fertilizer (the same fertilizer and the same dose as in experiment II).

Experiment IV: Evaluation of Ovine Manure, Municipal Solid Waste Compost, Sewage Sludge Compost and Olive Mill Pomace Compost as Fertilizers for Pine Replanting

Sixty homogeneous pine plantlets, of around 20 cm high, were grown individually in 10.5-liter plastic pots filled with forest soil (pH in soil: water dilution 1:2.5: 8.52; electric conductivity and Cl^- concentration in soil: water dilution 1:5: 0.306 dS m^{-1} and 26.2 mg l^{-1}, respectively; oxidable organic matter: 8.40 g kg^{-1}). Six fertilizer treatments (6 replicates each) were tested: C (non-amended soil), OM, MSWC, SSC, OMPC, MSWC/SSC (1:1), all the fertilizers were applied at the rate of 0.6 g N l^{-1}, the equivalent to the usual application of organic fertilizers in forest replanting.

Plant Measurements and Analytical Methods

At the end of the study (7 months), plant measurements and foliar analyses were done for all the experiments. The physico-chemical characteristics of the substrates and soils used in experiments III and IV, respectively, were also determined.

Plant height from the root collar to the terminal bud, stem diameter at the collar, as well as root, aerial and total plant dry weights, were used to calculate different morphological attributes: the Dickson Quality Index (DQI; Dickson et al., 1960), Sturdiness Quotient (SQ) and the Shoot/Root Ratio (SRR), according to the following formulae (Thompson, 1985):

$$SQ = \frac{H}{D}$$

$$SRR = \frac{ADW}{RDW}$$

$$DQI = \frac{TDW}{(SRR + Q)}$$

where H is plant height (cm), D is stem diameter (mm), and ADW, RDW, and TDW are the dry weights (g) of the aerial part, roots and the whole plant, respectively.

Analytical determinations were made in soils, substrates and foliar samples following the Official Methods of the Spanish Ministry of Agriculture, Food and Fisheries (MAPA, 1994), or with slight modifications. Electrical conductivity (EC) and Cl^- concentration were determined in aqueous solution 1:5 by a CyberScan CON 500 conductimeter (Eutech Instruments, MA, USA) and a PCLM3 chloryde analyzer (Jenway, Essex, UK), respectively; pH was measured with a pH-meter Basic 20 (Crison, Barcelona, Spain) in an aqueous solution 1:2.5 for soils and 1: 25 for organic refuse and substrates. Oxidized organic matter was determined by the Walkley-Black method (Walkley and Black, 1934) and nitrogen content by Kjeldahl digestion (Bremmer and Mulvaney, 1982) using a 2400 Kjeltec AutoSampler System (Foss Tecator AB, Sweden). Macro- and micronutrient concentrations were measured by simultaneous inductively coupled plasma atomic emission spectrometry (ICAP-AES 6000, Thermo Scientific, Cambridge, UK; Isaac and Johnson, 1998) after nitric-perchloric digestion (Campbell and Plank, 1998).

Data were subjected to an ANOVA to test for significant differences between treatments. Before carrying out any statistical analysis, the ANOVA assumptions were checked. The significance of the comparisons made among treatments was analyzed by Fisher's least significance difference (LSD) test at $P < 0.05$. All the statistical calculations were made using the Statgraphics Plus 5.0 (Manugistics Inc.) software package.

RESULTS AND DISCUSSION

Experiment I: Evaluation of Sewage Sludge Compost and Meat and Bone Meal as a Fertilizer for Nurseries

Two parameters that are normally employed for plant seedling quality are plant height and total dry weight. Neither the use of organic amendments or mineral fertilization nor considered N rate showed any significant differences for these characteristics (**Table 2**). The increase in heights varied between 11.7 cm in SSC2 to 16.6 cm in SSC1. The total dry weights ranged from 6.38 g in SSC2 to 9.24 g in C, with no differences found when comparing aerial and root weights.

Despite the fact that the previous parameters indicate growth, they are not related with the plant's survival capacity (Mañas et al., 2009). SQ, for example, indicates the mechanical resistance of trees and their capacity to support windy periods in the field. In this experiment, SQ showed no differences according to fertilization, with values varying between 5.85 in C and 6.25 in MBM1 (**Table 2**), which agree with those found for *Pinus* seedlings under Spanish nursery conditions (Domínguez-Lerena et al., 2001; Mañas et al., 2009).

Likewise, SSR is related with coniferous seedlings' capability to establish in arid and semiarid regions (South et al., 1985) and although it was not affected by treatment, its values varied between 2.34 in SSC1 and 2.86 in MBM1 (**Table 2**), which are clearly higher than the recommended value of 1 for *Pinus halepensis* (Domínguez-Lerena et al., 2001). Therefore, these plants could pose problems when being replanted in dry areas because their root development will not suffice to supply the water needs of their foliage biomass.

DQI is a global parameter that integrates total plant mass, SQ and the SRR ratio, and expresses the equilibrium between mass distribution and sturdiness. It avoids selecting disproportionate plants and refusing short, but vigorous, plants. The obtained values are independent of treatment, but come close to 1 (from 0.802 in SSC2 to 1.13 in C). So regarding this index, seedlings can be considered to be of good quality.

All the treatments met the nutritional plant requirements, as shown by the adequate cation contents in the foliar analyses (**Table 3**). Regarding N, only the seedlings amended with MBM2 presented similar levels to those with mineral fertilization. The other treatments had lower N concentrations, but gave values that fell within the normal limits reported for *P. halepensis* (Fürst, 1997 cited in Tausz et al., 2004). Different P inputs did not produce significant differences in foliar contents; K

TABLE 2 | Growing parameters and morphological quality index in fertilization assay.

Treatment	H (cm)	HI (cm)	AW (g)	RW (g)	TW (g)	SQ	SRR	DQI
C	29.7a	14.0a	6.75a	2.49a	9.24a	5.85a	2.78a	1.13a
SSC1	32.1a	16.6a	5.83a	2.46a	8.29a	6.06a	2.34a	1.05a
SSC2	27.3a	11.7a	4.62a	1.76a	6.38a	5.87a	2.60a	0.802a
MBM1	30.6a	15.1a	6.40a	2.24a	8.64a	6.25a	2.86a	0.959a
MBM2	30.4a	14.1a	5.44a	2.11a	7.55a	5.95a	2.53a	0.977a

HI, height increase. All weights are "dry weight." For each column values followed by the same letter do not differ significantly (LSD test, $p < 0.05$).

TABLE 3 | Average foliar macronutrient (N, P, K, Ca, and Mg; g Kg^{-1} dry matter) and micronutrient (Fe, Mn, and Zn; mg Kg^{-1} dry matter) content in fertilization assay.

Treatment	N	P	K	Ca	Mg	Fe	Mn	Zn
C	14.7b	1.27ab	18.0c	7.19c	2.38a	116b	25.2b	55.2b
SSC1	13.2a	1.28ab	15.7a	6.20ab	2.37a	95.5a	19.5a	47.2a
SSC2	13.6a	1.20ab	15.2a	6.04a	2.33a	106ab	42.6c	56.7b
MBM1	13.7a	1.29b	16.9b	6.64b	2.68b	90.5a	19.1a	45.6a
MBM2	14.7b	1.17a	16.6b	6.56ab	2.42a	87.2a	18.6a	43.9a
Adequate ranges	10–20	1–2	>8	3–6	1–1.9	20–200	20–800	20–70

For each column values followed by the same letter do not differ significantly (LSD test, $p < 0.05$).
Adequate ranges: macronutrients cited in Tausz et al. (2004) for P. halepensis; micronutrients in Fürst (2013) for Pinus spp.

concentration was, however, the lowest in the treatments including sewage sludge compost, which contained 4-fold less K than meat and bone meal. Ca levels were slightly high, probably due to the presence of this cation in irrigation water. No trace elements reached phytotoxic levels (**Table 3**) as concentrations fell within the ranges considered normal for the *Pinus* genus (Fürst, 2013).

We considered that all the seedlings presented characteristics according to growth and morphological standards, independently of fertilization type. Although significant differences in some elements were found depending on treatment, nutrient status was optimal in all cases. Organic fertilization thus led to plantlets with a similar quality to those fertilized with mineral products.

Experiment II: Evaluation of Vermicompost as a Root-Growth Promoter for Pine Plantlets

Plant growth was independent of vermicompost application: height increase and plant biomass did not vary with treatment (**Table 4**). No biomass radical stimulation was detected with the weights that ranged between 2.12 g in VC4 and 2.61 g in VC3. Morphological attributes were not affected: SQ fluctuated between 5.83 in VC1 and 6.37 in VC4; SRR between 2.71 in C and 3.08 in VC4; DQI between 1.01 in VC4 and 1.15 in VC3 (**Table 4**).

The concentration of nutrients in leaves showed significant differences between treatments, but no clear trend was concluded when comparing the applied rate and the control (**Table 5**). Vermicompost amendments did not, thus, improve the nutritional state of plants. In general, the macro- and micronutrient contents in leaves can be considered optimal, but Ca and Mg concentrations were slightly high and totally independent of treatment.

Although vermicompost has been related to enhanced plant growth, radical development and nutrient uptake according to different studies, no effect was seen in our assay, not even at the highest dose. Lazcano et al. (2010) found no stimulation of aerial or root biomass after vermicompost addition in *P. pinaster* seedlings; conversely, the N content in leaves increased. The NPK fertilization applied in our study could have masked this effect. Nor did we rule out the possibility of another kind of vermicompost having positive effects as this matter's chemical and biological properties can widely vary depending on the initial organic waste (Madan et al., 2009), the production process (Pramanik et al., 2007), and even on the earthworm species used (Lores et al., 2006) or the predominant age group (Fu et al., 2014). Some authors have found contrasting effects of vermicompost depending on plant species, even among varieties of the same species (Lazcano et al., 2010).

Experiment III: Evaluation of Coconut Fiber, Recycled Coconut Fiber, Municipal Solid Waste Compost and Ground Olive Stone as Forestry Substrate Components

The main characteristics of the evaluated substrates are shown in **Table 6**. Commercial customized substrate presented the highest pH, 9.19, and the other substrates came close to neutrality. Electric conductivity (EC) varied from 2.35 dS m^{-1} in the control to 6.98 dS m^{-1} in RCF/MSWC 2:1, and was higher in the mixtures with compost. Although salt concentrations exceeded adequate values ($\leq$0.5 dS m^{-1}, Abad et al., 2001) in all cases, plant injury did not occur. Other authors have noted this effect when using such substrates (Hernández-Apaloaza et al., 2005). The high salt-buffering capacity of the materials, because of their organic nature, probably protects the root system, or the irrigation and the consequent medium leaching lowers salt concentrations to acceptable levels.

Bulk densities were lower than 0.4 g cm^{-3} in all cases, except in substrate C. This density is the maximum recommended value for adequate root elongation and easy substrate manipulation (Abad et al., 2001). The lowest bulk density went to CF, 0.0900 g cm^{-3}, but agrees with the normal range for such materials (Abad et al., 2005). Water-holding capacities also had the recommended value of 60–100% (Abad et al., 2001).

No significant differences in height increase were observed among the different treatments (**Table 7**). Values varied between 13.1 cm in the plants grown in RCF/MSWC 3:1 and 22.7 cm in those grown in CF. The plants in coconut fiber obtained a significantly higher total weight value (CF: 16.7 g, RCF: 13.9 g) than those grown in customized commercial substrate (8.84 g) due to major aerial biomass development (**Table 7**). No significant differences were found between using virgin or recycled coconut fiber. Recycled coconut fiber either alone or combined with municipal solid waste compost or ground olive stone gave plants with similar total weights, except for the RCF/MSWC/GOS 2:1:1 combination as plants were lighter.

TABLE 4 | Growing parameters and morphological quality index in vermicompost assay.

Treatment	H (cm)	HI (cm)	AW (g)	RW (g)	TW (g)	SQ	SRR	DQI
C	31.8a	14.8a	6.87a	2.59a	9.46a	6.25a	2.71a	1.07a
VC1	31.6a	15.0a	7.22a	2.47a	9.69a	5.83a	2.89a	1.10a
VC2	33.0a	16.2a	6.79a	2.28a	9.07a	5.93a	3.03a	1.02a
VC3	31.0a	14.0a	7.33a	2.61a	9.94a	6.03a	2.84a	1.15a
VC4	32.4a	15.6a	6.39a	2.12a	8.51a	6.37a	3.08a	1.01a
VC5	32.4a	14.8a	7.46a	2.60a	10.1a	6.36a	2.90a	1.08a

HI, height increase. All weights are "dry weight." For each column values followed by the same letter do not differ significantly (LSD test, $p < 0.05$).

TABLE 5 | Average foliar macronutrient (N, P, K, Ca, and Mg; g Kg^{-1} dry matter) and micronutrient (Fe, Mn, and Zn; mg Kg^{-1} dry matter) content in vermicompost assay.

Treatment	N	P	K	Ca	Mg	Fe	Mn	Zn
C	15.0b	0.920a	18.1c	7.90b	2.50cd	107b	23.4a	58.3c
VC1	14.4a	0.970ab	15.8a	7.10a	2.40bc	88.0a	30.8b	48.6bc
VC2	14.5ab	0.980ab	17.6bc	7.20a	2.30b	105b	24.0a	50.5bc
VC3	14.9b	0.950ab	16.8abc	8.20b	2.60d	111b	28.6ab	54.6bc
VC4	14.2a	1.02b	16.8abc	7.90b	2.50cd	117b	24.0a	47.7a
VC5	14.2a	1.01b	16.5ab	6.70a	2.10a	111b	40.1c	48.2a
Adequate ranges	10–20	1–2	>8	3–6	1–1.9	20–200	20–800	20–70

For each column values followed by the same letter do not differ significantly (LSD test, $p < 0.05$).
Adequate ranges: macronutrients cited in Tausz et al. (2004) for P. halepensis; micronutrients in Fürst (2013) for Pinus spp.

TABLE 6 | Physico-chemical characteristics of the substrates.

Treatment	pH (1:25)	EC (dS m^{-1})	Cl^- (mg l^{-1})	BD (g cm^{-3})	WHC (%)
C	9.19	2.35	641	0.467	54.5
CF	5.44	3.73	207	0.0900	67.8
RCF	6.18	4.41	304	0.120	72.2
RCF/MSWC 2:1	7.26	6.98	1,137	0.180	67.4
RCF/MSWC 3:1	7.02	6.48	931	0.160	67.2
RCF/GOS 2:1	6.84	3.13	209	0.273	61.9
RCF/GOS 3:1	6.77	3.62	233	0.233	63.9
RCF/MSWC/GOS	7.37	4.77	634	0.293	60.5

EC, electric conductivity in soil: water dilution 1:5; BD, bulk density; WHC, water holding capacity.

Quality parameters showed no major differences among treatments, and their values fell within the range obtained in the other experiments carried out. The SRR values were high in the treatments with only coconut fiber (CF and RCF) because aerial growth was significantly enhanced. The virgin coconut fiber obtained the worst DQI value.

As **Table 8** indicates, all the plants presented adequate N, K and micronutrient contents. Although P, Ca and Mg slightly exceeded the recommended values in certain treatments, plantlets generally manifested an acceptable nutrient status.

Our results showed that the plants grown in virgin or recycled coconut fiber, and in combination with recycled coconut fiber and MSWC or GOS, were generally similar to those grown in the commercial substrate. Indeed some parameters like plant weight or pH and bulk density of the substrate actually improved. The customized commercial substrate normally employed in *P. halepensis* seedlings in the Mediterranean Region can be substituted for other organic substrates, mainly for the combinations of recycled coconut fiber with MSWC or GOS, as they produce plantlets with a better aerial and radical biomass balance. Previous studies have reported the feasibility of using organic waste as substitutes for the substrates commonly used in forest nurseries, but variable results have been obtained depending on plant species and organic waste characteristics. Hernández-Apaloaza et al. (2005) improved the growth of Cupressus sempervirens and C. arizonica seedlings using coconut fiber with 30% (v/v) of SSC, but detected no effect on *P. pinea* seedlings. Guerrero et al. (2002) reported an increase in plant biomass in C. arizonica by substituting peat for pine bark and sewage sludge mixtures, but obtained no response in *P. pinea*. Mañas et al. (2009) reported positive results in P. pinaster seedlings when peat was partially substituted for organic waste, but different effects were found depending on the characteristics: mixtures of paper mill sludge with activated sewage sludge gave the highest physical parameter values (height, root-collar diameter, total dry weight), while mixtures of SSC with MSWC generated plants with the best SQ and SRR values.

Use of Sheep Manure, Municipal Solid Waste Compost, Composted Sewage Sludge and Composted Olive Mill Solid Residue as Fertilizers in Reforestation Pines

One of the main problems of Mediterranean soils is their low organic matter content. Consequently, using organic amendments is crucial in afforestation programs. In this experiment, different locally available organic waste types were evaluated as fertilizers when pines were replanted in soil. As **Table 9** shows, amendments clearly increased soil organic matter. The lowest value was for the non-amended soil with only 8.40 g kg^{-1}, while SSC and MSWC, with the highest contents, 22 and 20.4 g kg^{-1}, respectively. The addition of organic

TABLE 7 | Growing parameters and morphological quality index in substrates assay.

Treatment	H (cm)	HI(cm)	AW (g)	RW (g)	TW (g)	SQ	SRR	DQI
C	33.3a	16.6a	6.50a	2.34a	8.84a	6.43a	2.80ab	0.964a
CF	39.2a	22.7a	12.8c	3.85a	16.7c	6.24a	3.46cd	1.84c
RCF	38.5a	22.1a	10.8bc	3.06a	13.9bc	6.60a	3.63d	1.38ab
RCF/MSWC 2:1	32.6a	16.2a	7.81ab	2.87a	10.7ab	5.61a	2.72ab	1.41bc
RCF/MSWC 3:1	29.1a	13.1a	8.41ab	2.75a	11.2ab	5.66a	3.09bc	1.32ab
RCF/GOS 2:1	33.6a	17.1a	8.25ab	2.92a	11.2ab	5.50a	2.82ab	1.53bc
RCF/GOS 3:1	37.4a	20.6a	8.69ab	3.52a	12.2ab	6.68a	2.55a	1.48bc
RCF/MSWC/GOS	33.5a	15.9a	6.73a	2.54a	9.27a	5.78a	2.76ab	1.39abc

HI, height increase. All weights are "dry weight." For each column values followed by the same letter do not differ significantly (LSD test, $p < 0.05$).

TABLE 8 | Average foliar macronutrient (N, P, K, Ca, and Mg; g Kg^{-1} dry matter) and micronutrient (Fe, Mn, and Zn; mg Kg^{-1} dry matter) content in substrates assay.

Treatment	N	P	K	Ca	Mg	Fe	Mn	Zn
C	13.8cd	1.20ab	14.5f	6.40d	2.30cd	109e	25.8a	36.0a
CF	15.9e	2.20c	11.4bc	5.30b	2.20bc	78.3a	97.8e	52.4cd
RCF	15.2e	3.00e	17.0g	6.40d	2.30c	93.8bc	72.8d	36.1a
RCF/MSWC 2:1	13.2bc	1.00a	9.60a	6.80e	2.20bc	95.6bc	29.4a	57.1d
RCF/MSWC 3:1	14.1d	1.40b	10.3ab	7.70f	2.40d	95.8bc	28.2a	46.4bc
RCF/GOS 2:1	13.6cd	2.60d	12.0cd	5.60c	2.10b	101bc	49.8c	37.4a
RCF/GOS 3:1	11.8a	3.10e	12.8ed	5.20b	2.20bc	91.3b	56.6c	39.7ab
RCF/MSWC/GOS	12.5ab	1.30b	13.5ef	4.90a	1.90a	108de	38.9b	56.2d
Adequate ranges	10–20	1–2	>8	3–6	1–1.9	20–200	20–800	20–70

For each column values followed by the same letter do not differ significantly (LSD test, $p < 0.05$).
Adequate ranges: macronutrients cited in Tausz et al. (2004) for P. halepensis; micronutrients in Fürst (2013) for Pinus spp.

amendments improves physical, chemical and biological soil properties (Querejeta et al., 1998, 2000; Larchevêque et al., 2006) and promotes trees establishment. In fact using native species supplemented with organic matter is the best restoration strategy, as recommended by the European Union (Caravaca et al., 2003). Except for sheep manure, all the treatments lowered pH value compared to the non-amended soil, especially composted sewage sludge. Garcia et al. (2000) noticed this effect when using organic amendments in *P. halepensis* afforestation, and attributed it to organic acids forming during the mineralization of the added organic matter.

The soils amended with composted sewage sludge obtained the highest values for EC and in Cl^{-} content, with 0.530 dS m^{-1} and 45.6 mg l^{-1}, respectively. A previous work conducted with digested or composted biosolid amendments has shown salinity problems associated with high contents in soluble salts (Cheng et al., 2007; Fuentes et al., 2007) but, as in our former assay into their use as substrates, plant injury did not occur.

The height increments at the end of the assay were not significant different between treatments, with values varying from 14.7 cm in OMPC to 17.3 cm in SSC (**Table 10**). Organic amendments commonly have positive effects on *P. halepensis* growth (Querejeta et al., 1998; Valdecantos, 2001; Fuentes et al., 2008), especially in relation to N and P availability. So if the levels of these nutrients are adequate in non-amended soil, treatments have no effect (Fuentes et al., 2010). Regarding biomass, the root dry weight in the control (6.93 g) was significantly higher compared to that in MSWC, OM and OMPC, with 4.85, 4.48 and 4.28 g, respectively. Apparently, the enhanced nutritional and structural conditions in soil caused by organic amendments induced less effort in plantlets to develop a strong and deep radical system, as noted by Valdecantos (2001) in *P. halepensis* and Vaitkute et al. (2010) in *P. sylvestris*. Morphological attributes did not significantly vary with amendments, with values that fell in line with those obtained in the previously performed assays.

TABLE 9 | Physico-chemical characteristics of the soils in reforestation fertilizers assay.

Treatment	OM_ox. (g Kg^{-1})	pH (1:2.5)	EC (dS m^{-1})	Cl^{-} (mg l^{-1})
C	8.40a	8.52c	0.306a	26.2a
OM	18.8bc	8.52c	0.370bc	24.2a
MSWC	20.4cd	8.41b	0.379bc	29.9a
SSC	22.2d	8.22a	0.530c	45.6b
OMPC	17.3b	8.51c	0.355ab	25.4a
MSWC/SSC	19.3bc	8.39b	0.399b	30.1a

OM_ox., oxidable organic matter; pH in soil: water dilution 1:2.5; EC, electric conductivity in soil: water dilution 1:5.

TABLE 10 | Growing parameters and morphological quality index in fertilization pine replanting assay.

Treatment	H (cm)	HI (cm)	AW (g)	RW (g)	TW (g)	SQ	SRR	DCI
C	37.8a	16.7a	14.8a	6.93c	21.7a	5.97a	2.19a	2.70a
OM	35.5a	15.3a	11.9a	4.48a	16.4a	5.94a	2.85a	1.91a
MSWC	35.3a	16.4a	12.9a	4.85ab	17.7a	6.13a	2.79a	2.08a
SSC	37.7a	17.3a	14.7a	6.55bc	21.2a	6.17a	2.26a	2.59a
OMPC	36.0a	14.7a	11.3a	4.28a	15.6a	6.56a	2.73a	1.80a
MSWC/SSC	34.5a	15.3a	12.3a	5.12abc	17.4a	6.07a	2.37a	2.08a

HI, height increase. All weights are "dry weight." For each column values followed by the same letter do not differ significantly (LSD test, $p < 0.05$).

TABLE 11 | Average foliar macronutrient (N, P, K, Ca, and Mg; g Kg^{-1} dry matter) and micronutrient (Fe, Mn, and Zn; mg Kg^{-1} dry matter) content in fertilization pine replanting assay.

Treatment	N	P	K	Ca	Mg	Fe	Mn	Zn
C	13.6a	1.00a	7.70a	6.91a	2.18a	171ab	37.9b	53.2a
OM	13.6a	1.10ab	11.0b	7.33a	2.54a	166ab	29.1a	51.0a
MSWC	14.1a	1.10ab	9.60ab	8.29a	2.49a	201ab	29.1a	65.1a
SSC	14.5a	1.40b	9.50ab	8.13a	2.36a	170ab	30.2a	53.4a
OMPC	13.7a	1.20ab	11.5b	6.88a	2.40a	248b	33.3ab	64.8a
MSWC/SSC	13.4a	1.10ab	8.70a	7.52a	2.45a	154a	27.8a	56.0a
Adequate ranges	10–20	1–2	>8	3–6	1–1.9	20–200	20–800	20–70

For each column values followed by the same letter do not differ significantly (LSD test, $p < 0.05$).
Adequate ranges: macronutrients cited in Tausz et al. (2004) for P. halepensis; micronutrients in Fürst (2013) for Pinus spp.

Trees presented adequate N, P and K contents, with a few or no significant differences between treatments and controls (**Table 11**). In pioneer species like *P. halepensis,* foliar concentration of the most limiting nutrients tended to remain at relatively constant levels when availability increased, and used extra inputs for growing (Bazzaz, 1979). Some authors have demonstrated this effect, like Fuentes et al. (2007) who noted stem height growth in the seedlings that received a biosolid treatment or Valdecantos (2001) who observed bigger needles instead of higher N contents. As in previous assays, all the treatments presented high Ca and Mg values (**Table 11**). Trace elements fell within a suitable range, except for Fe in the OMPC treatment as it slightly exceeded the recommended level. Nevertheless, seedlings in general displayed adequate nutrient conditions.

Sheep manure is the product that is normally added in the Mediterranean Region while transplanting. In this assay, no significant differences were found according to amendment in either soil characteristics or tree quality. This finding supports the possibility of substituting sheep manure for other refuse materials. Organic waste of different origins have already been reported as suitable afforestation amendments, like municipal waste composts and different by-products from the olive oil industry, thanks to their noticeable fertilizing capacity (Garcia et al., 2000; Caravaca et al., 2003; Murillo et al., 2005).

CONCLUSIONS

Although this is an exploratory study, the results indicate that locally accessible organic waste and composts in the Mediterranean Region can be used for different purposes in *P. halepensis* seedlings to avoid using non-renewable materials. Organic fertilization in nurseries with SSC and MBM gave similar results to mineral fertilization, with no effect on the considered N rate. All the treatments met the nutritional plant requirements and plantlets reached quality standards. The addition of OM, MSWC, SSC, OMPC and MSWC/ SSC (1:1) as fertilizers for plant replanting clearly increased soil organic matter. Although the trees amended with OM, MSWC and OMPC presented less root development than those grown in the non-amended soil, treatments did not differ in terms of height, morphological attributes and nutritional status. These results support the possibility of substituting sheep manure, usually employed for replanting in the Mediterranean Region, for other locally available organic refuse. When evaluating the different coconut fiber mixtures, MSWC and GOS as substrates, plantlets were generally similar to those grown in commercial substrate, and the RCF/MSWC (2:1 and 3:1 v/v) and RCF/GOS (2:1 and 3:1 v/v) combinations obtained a better balance in terms of their aerial and radical biomass. Conversely, vermicompost was not useful for stimulating root development at any assayed dose, nor were plant growth, morphological parameters and nutrient content affected by its application.

AUTHOR CONTRIBUTIONS

RC-C and RA designed the experiment and supervised the experimental work; RM and MR performed the experimental work and the statistical analysis; RM and AP-P contributed to the discussion of the results; ML-M wrote the first draft of the manuscript; AP-P wrote the final version of the manuscript. All authors approved the submitted version.

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